

VRBPAC January 31, 2001
LYMERix® Lyme Disease Vaccine, safety update

Introduction

LYMERix®, Lyme Disease Vaccine, manufactured by SmithKline Beecham Pharmaceuticals, is indicated for the prevention of Lyme disease in subjects 15-70 years of age. The vaccine is composed of 30 mcg recombinant lipidated OspA with alum. OspA is the major outer surface protein of the spirochete, *Borrelia burgdorferi*, the etiologic agent of Lyme disease.

In 1993, the Sponsor began their initial studies of recombinant OspA as a human antigen in Europe. In 1994, a US IND was initiated and the sponsor began a dose ranging safety and immunogenicity study. In June 1994, VRBPAC met to discuss the clinical design of Phase 3 studies intended to evaluate the safety and efficacy of Lyme disease vaccines. The sponsor initiated the Phase 3 efficacy study in January 1995. Results of this study were submitted to the FDA as part of a Product License Application on September 15, 1997, and presented to VRBPAC on May 26, 1998. FDA licensed LYMERix® on December 21, 1998.

The safety and efficacy data that formed the basis for the approval of LYMERix® are summarized in the Summary Basis of Approval; a copy of this document is provided with the briefing package.

Prior to licensure of the LYMERix® recombinant OspA vaccine, several research reports and epidemiological studies suggested that either anti-OspA antibodies or OspA-specific T cells, particularly in people who have DR4 major histocompatibility (HLA) alleles, were associated with the development of treatment resistant Lyme arthritis (TRLA). TRLA is a rare syndrome in which as many as 10% of persons who experience arthritis as a late clinical manifestation of Lyme disease have persistent symptoms for months or even several years after antibiotic treatment (Steere *et al.*, Arthritis and Rheumatism 37: 878-888, 1994). Research into the pathogenesis of TRLA is ongoing, and invokes concepts about the fundamental nature of antigen recognition by T lymphocytes that are still incompletely understood. In 1998, one research group hypothesized that TRLA was a consequence of T cell recognition of a particular peptide of the *B. burgdorferi* OspA via presentation by particular HLA alleles. The HLA types in question included the so-called rheumatoid arthritis-associated or "RA" alleles, exemplified by DRβ1*0401; many of these share a sequence in the third hypervariable region of the DRβ1 chain. Further, the authors hypothesized that the human cell surface adhesion molecule Lymphocyte Function Antigen 1 (LFA-1), which is expressed on activated T cells, contained a homologous peptide that could continue to stimulate T cell activation after elimination of the spirochete. In this hypothesis, the exact mechanism by which chronic T cell stimulation and (Th1) cytokine secretion would actually lead to immunopathology and frank arthritis was not specified.

Data presented in support of this hypothesis (Gross *et al.*, Science 281: 703-706, 1998) included the demonstration that T cells from synovial fluid for 14 of 16 TRLA patients (but 5 of 16 patients with treatment responsive Lyme arthritis, or 3 of 9 control arthritis patients) produced interferon γ, and/or proliferated, in response to whole OspA. Synovial T cells also responded to a particular immunodominant peptide of OspA (amino acids 164-175), and a

recently developed binding algorithm predicted binding of this OspA₁₆₄₋₁₇₅ peptide to the HLA DRβ1 0401 molecule. Further, a peptide from LFA-1 had a 6 amino acid homology with the OspA peptide as well as a high score in the binding algorithm; direct binding of the LFA-1 peptide to DRβ1 0401 was demonstrated *in vitro*, implying the potential for autoimmune cross-reactivity between OspA and LFA-1. Data was also presented indicating that synovial T cells from two TRLA patients produced increased interferon γ in response to OspA, the OspA peptide, human LFA-1, and the LFA-1 peptide.

On the other hand, other data presented in this and subsequent publications do not necessarily support this hypothesis. For example, cells from several TRLA subjects studied did not appear to recognize either OspA, the OspA peptide, LFA-1, or the proposed cross-reactive LFA-1 peptide (despite having one of the relevant RA alleles), and some responses were quite low or spontaneous cytokine release without stimulation quite high (Gross *et al.*, Science 281: 703-706, 1998). It should also be noted that relatively few subjects have been studied to date, and that the studies reported have not been confirmed by other investigators. In follow up studies from the same group, only T cells from synovial joint fluid were reactive, while peripheral blood T cells from the same patients were not (Chen *et al.*, Arthritis Rheum. 42: 1813-1822, 1999). When T cells specific for the OspA₁₆₄₋₁₇₅ peptide were cloned from the synovial fluid of 3 TRLA patients, only a small fraction of these clones bound an MHC DRβ1 0401 tetramer loaded with the LFA peptide (Meyer *et al.*, PNAS 97: 11433-38, 2000), and only 11% proliferated in response to human LFA-1 protein (SKB CMI report; see below). The latter findings highlight the uncertainty of the LFA-1 cross-reaction at this time.

Recent studies from other investigators also suggest alternate interpretations of the data concerning T cell responsiveness in TRLA patients. Investigations using a combinatorial peptide library to screen a *B. burgdorferi*-specific T cell clone derived from a patient with chronic neuroborreliosis found not only several *Borrelia* antigens that could stimulate proliferation, but also several other potential autoantigens that had minimal (or no) obvious sequence homology with the putative bacterial antigens (Hemmer *et al.*, Nature Med. 5: 1375-1382, 1999). Such results imply that structural homology may be neither necessary nor informative, and that T cell recognition may have flexibility and plasticity that is different in different environments. Similarly, another group used peptide spot synthesis to perform a complete peptide substitution analysis of antigens stimulating T cell hybridomas derived from OspA-immunized DR4 transgenic mice, including hybridomas responsive to the OspA₁₆₄₋₁₇₅ peptide. Binding motifs identified by these analyses were used to screen databases for matching human or mouse peptides, and these candidate autoantigens were again used to test stimulation of the original clones. Several dozen mammalian proteins with apparent cross-reactivity were readily identified by this approach, suggesting that T cell cross-reactivity may actually be quite common (Maier *et al.*, Eur. J. Immunol. 30: 448-457, 2000); if so, the existence of such cross-reactions alone would not imply a molecular mimicry outcome, as autoimmunity is obviously uncommon.

The hypothesis discussed above concerns the role of OspA and OspA-responsive T cells in the pathogenesis of TRLA, which follows infection with *Borrelia burgdorferi*. It is unclear whether there is any relationship between this complication of Lyme disease itself (as initiated by infection with a living, motile pathogenic bacterium), and vaccination with purified OspA

protein. Nonetheless, the clinical trials investigating safety and efficacy of LYMERix® were prospectively designed to examine whether vaccinees experienced higher incidences of rheumatological or neurological symptoms than placebo recipients. No increased incidence was observed. Of note, it likely that about 30% of the individuals enrolled in the clinical efficacy trial carried a DR4 allele, and approximately 5% an RA allele.

SmithKline Beecham also incorporated an exploratory research study of cell mediated immune (CMI) responses to OspA in LYMERix® vaccinees in the pivotal efficacy trial, and licensure of LYMERix® included a commitment to provide a final report of this study to FDA. This report has been received. In this study, peripheral blood lymphocytes (PBLs) obtained from about 50 vaccinee and 50 placebo subjects were HLA typed and studied for proliferation and cytokine secretion following stimulation with OspA or the OspA₁₆₄₋₁₇₅ peptide. PBLs from 13/41 (32%) vaccinees proliferated in response to whole (unlipidated) OspA, and 9 of the 13 had a “positive” proliferative response to the OspA₁₆₄₋₁₇₅ peptide. However, responses to the peptide were very low, generally much lower than the response to whole OspA protein. Not surprisingly, cells from only 1/44 of the placebo subjects responded. Cytokine (IFN γ and IL4) responses to either whole OspA or any of the peptides were uniformly very low, with only a few sporadic marginal positives. Although 30% of the vaccinees had peripheral blood CMI responses to OspA, these responses were low, and the responses to the OspA₁₆₄₋₁₇₅ peptide almost undetectable. These results were also analyzed by stratifying vaccinees into 3 groups according the presence of DR β 1*0401 /*0404 ((RA alleles), 6 subjects; DR β 1*11 (no increased risk of TRLA), 10 subjects; or other DR alleles (mixed, possibly some risk with some alleles but only low resolution typing performed), 25 subjects. No trends were apparent, especially as responses were low and numbers of subjects small. There were also no obvious correlations between adverse events in any of these subjects and either OspA responsiveness or HLA type. Finally, reactivity to LFA-1 or the LFA-1 peptide was not studied; thus these studies neither support nor refute the hypothesis concerning OspA and LFA-1 cross reactivity.

Also of note, with possible implications for the safety of LYMERix®, are recent reports from a number of laboratories suggesting that long recognized mitogenic properties of OspA may be due to the ability of OspA (and other bacterial lipoproteins) to initiate signal transduction, lymphocyte activation, and inflammatory responses by engaging the newly described family of cell surface receptors known as Toll-like receptors (TLR). *In vitro* induction of IL-12 secretion by macrophages (Brightbill *et al.*, Science 285: 732-736, 1999), activation of NF κ B (Hirshfeld *et al.*, J. Immunol. 163: 2382-2386, 1999; Lorenz *et al.*, Infect. Immun. 68: 6398-6401, 2000), and stimulation of TNF α production (Lien *et al.*, J. Biol. Chem. 274: 33419 - 33425, 1999) have been reported to date. At the present time, the relevance of these *in vitro* studies to vaccination, and any implications for the dose of OspA used, are not clear.

At the time of licensure, the original PLA submission contained safety data regarding the administration of 18,047 doses of the of OspA Lyme disease vaccine (30 mcg) to 6,476 subjects who were ≥ 15 years of age. For details of the safety analyses refer to the SBA. While exposure to LYMERix® was associated with significant increases in transient post-injection symptoms, no increase was seen in the incidence of any chronic musculoskeletal events, and in the incidence of any conditions associated with chronic Lyme disease when vaccinees were compared to control. It was noted that subjects with a prior history of

musculoskeletal conditions and those with a self-reported history of Lyme disease were more likely to experience musculoskeletal events than subjects without such prior history, but there were no differences in incidence of musculoskeletal events when placebo and vaccine-recipients were compared within any of these groups.

However, because of theoretical concerns about the association of anti-OspA antibodies and limitations of prelicensure studies in detecting rare adverse events, the committee recommended that additional post-licensure studies be performed to further evaluate the safety profile of LYMERix®. At the time of approval, SmithKline Beecham agreed to conduct a Phase 4 post-marketing study in 25,000 vaccinated individuals (a synopsis of the study is provided in the briefing document), to establish a pregnancy registry to specifically monitor adverse events in pregnant women, to complete a preclinical reproductive toxicity study, and to complete a cellular immunity study.

Since licensure of LYMERix®, reports of adverse events occurring following receipt of the vaccine have circulated in the media. The purpose of today's meeting is to review the safety database, which supported the licensure of LYMERix®, to update the committee on the status of post-marketing studies and adverse events reported to the VAERS system. (A summary of the VAERS data is provided with the briefing document.)